REMARKS

The abstract and specification have been amended in order to correct grammatical and idiomatic errors contained therein. No new matter has been added.

The claims have been amended in order to respond to the Examiner's rejections under 35 USC 112 and to more particularly point out and distinctly claim the subject matter which Applicants regard as the invention. Newly presented Claims 7-15 are directed to preferred embodiments of the present invention. No new matter has been added.

Claims 1-3 have been rejected under 35 USC 112, first paragraph, as not providing enablement or a method of treating a tumor with any SOD. Claims 1-6 also have been rejected under 35 USC 112, first paragraph, for not providing enablement for a method of preventing a tumor by the administration of treated SOD, protein or lipid-treated SOD, SOD combined with gliadin or melon SOD combined with gliadin. Applicants respectfully submit that the currently presented claims clearly comply with 35 USC 112, first paragraph.

With respect to the Examiner's first rejection under 35 USC 112, first paragraph, Applicants point out that SOD is an enzyme having a specified activity regardless of its source. Although SODs from different sources may have different enzymatic activity, the currently presented claims require that a pharmacologically effective amount of the SOD be administered. Therefore, although only one specific SOD is tested in the present specification, this by no means limits the scope of the present invention to the tested SOD as one of ordinary skill in the art would be readily aware that the currently presented invention applies to any SOD, irrespective of its source.

With respect to the rejection of Claims 1-6 under 35 USC 112, first paragraph, as discussed in the Background of the Invention in the present specification at page 2, line 25, a new method of evaluating malignant progression was used to

study the anti-tumor activity of melon SOD coated with gliadin and it was found that this composition prevented malignant transformation of benign tumor cells to malignant tumor cells as shown in Result 2 of the Example on page 8 of the present specification. These results showing that SOD-G prevented the progression or malignant transformation of benign tumor cells indicates that SOD-G has a tumor-preventing ability as discussed on specification page 1, lines 14-27, and specification page 5, lines 11 and 12. As such, it is respectfully submitted that Claims 1-6 clearly comply with the requirements of 35 USC 112, first paragraph.

Claims 1 and 2 have been rejected under 35 USC 112, second paragraph, as being indefinite. This rejection has been overcome by the incorporation of Claim 3 into Claim 1. Accordingly, Claims 2 and 3 have been canceled. Newly presented Claims 7-15 are directed to preferred embodiments of the present invention. It is respectfully submitted that the currently presented claims clearly comply with the requirements of 35 USC 112.

Claims 1 and 2 have been rejected under 35 USC 102(b) as being anticipated by Ginoux et al. Claims 1-6 have been rejected under 35 USC 103(a) as being unpatentable over Murcia et al in view of Postaire et al and Ginoux. Applicants respectfully traverse these grounds of rejection and urge reconsideration in light of the following comments.

The presently claimed invention is directed to a method of preventing or treating a tumor which comprises the step of administering to a subject in which the tumor is to be treated or prevented a pharmacologically effective amount of superoxide dismutase combined with gliadin. The present invention is based on the discovery that the treated SOD of the present invention functions as an induction agent for the host antioxidant enzymes including SOD, catalase and glutathion peroxidase and is therefore essentially different from conventional SOD preparations which do not effect the SOD levels at any tissues and acts only by itself as an

antioxidant at the site of the drug administration. That is, the treated SOD of the present invention was tested for its ability to inhibit tumor growth and malignant progression through the use of the inflammatory cell mediated progression model in mice and it was discovered that SOD-G inhibited tumor growth and malignant progression.

Orgotein is a purified protein of superoxide dismutase obtained from bovine liver and red blood cells and is an anti-inflammatory drug developed in Europe as is disclosed in the enclosed Reference 1. Orgotein is not administered orally because, as shown in the enclosed Reference 2, it has been reported that the oral route of administration of Orgotein did not elevate tissue levels of SOD activity. Chemical investigation of Orgotein showed that Orgotein was more effective in patients who had developed an antibody against bovine SOD during repeated injections of the drug. Accordingly, in the enclosed Reference 3, the function of specific antibodies against bovine SOD was studied and it was found that bovine SOD and its specific antibody complex carried SOD into Murine macrophages. The intracellular localization of bovine SOD-anti-bovine SOD antibody complex into the Murine macrophages induced higher levels of production of Murine antioxidant enzymes including SOD, catalase, glutathion peroxidase and glutathion as illustrated in Table 5 of Reference 3. As a control, bovine SOD alone did not induce any of the antioxidant enzymes or glutathion.

There are three types of anti-oxidant enzymes produced to detoxicate the toxicity of reactive oxygens. These are SOD, catalase and glutathion peroxidase. Reference 1, discussed above, disclosed that all three kinds of anti-oxidant enzymes, which are the gene products of host DNA, are induced by bovine SOD-anti-bovine SOD immune complex, and, in the patients who showed good clinical efficacy of Orgotein, the specific antibody against Orgotein is the key molecule in inducing higher levels of the three antioxidant enzymes and glutathion, which results in good clinical efficacy. In light of this

knowledge, the references cited by the Examiner will now be discussed.

The Ginoux et al reference discloses a protein extract from *Cucumis Melo* having an elevated superoxide dismutase enzyme activity. This protein extract is disclosed as being useful for cosmetic purposes, medical purposes, food purposes and contains, as an active ingredient, an optionally purified protein extract.

At the outset, the Ginoux et al reference does not disclose the combination of the superoxide dismutase with a gliadin. As such, the presently claimed invention clearly is patentably distinguishable over this reference. Additionally, as a pharmaceutical agent, this reference discloses the use of the protein extract in the treatment of certain cancers of the digestive system. This is because orally administered SOD can contact directly with tumors located in the digestive tract before inactivation of the SOD occurs. Typically, proteinase in the digestive juice destroys SOD activity. This means that if any anti-tumor effect is exhibited, it would be effective only in the digestive system which comes into direct contact with the SOD. As opposed to this reference, the present invention discloses that the treated SOD is effective on a subcutaneously implanted mouse tumor, liver metastasis as well as prostate cancer. As such, the treated SOD of the present invention is an effective anti-tumor agent against a tumor located anywhere in the subject's body.

It has previously been thought that SOD inhibits the anti-cancer activity of x-ray irradiation, anti-cancer drugs and anti-tumor activity of immune leukocytes because reactive oxygen generated by x-ray irradiation, various anti-cancer drugs such as Mitomycin C, Adriamycin, Cisplatin, Bleomycin and others, and macrophages in NK cells kill cancer cells by releasing reactive oxygen as disclosed in the enclosed Reference 4. In fact, the enclosed Reference 5 reports that an increase of SOD in cancer cells diminishes the cytotoxic effect of several anti-cancer modalities and inhibition of SOD

activity augmented anti-cancer activity of radiation and some anti-cancer drugs.

The "treated" SOD of the present invention was prepared in an 80% ethanol solution as described in the present specification at page 6, lines 13-22. Methanol and prolamines are not contained in melons. As such, it is impossible to form a treated melon SOD spontaneously and, in the case of a lipid-treated SOD, the SOD must be incorporated in a liposome. Liposome formation is produced under a specific artificial condition of lipid constitution which requires a mixture of a phospholipid and cholesterol. Therefore, it is respectfully submitted that the treated SOD of the present invention is not produced spontaneously in the crude extract of a melon, even if lipid materials are contained in the melon extract.

The Murcia et al reference discloses that melons, among other Mediterranean and tropical fruit, have antioxidant activity. However, there is no disclosure in this reference regarding SOD activity in that the Murcia et al reference studied the antioxidant or scavenger activity of melon pulp homogenate by estimating activities against the hydroxyl radical, HOCl and hydrogen peroxide. No activity against superoxide dismutase was measured because SOD activity is measured only when dismutation activity of O_{2^-} is measured. Applicants admit that there is SOD present in melons because all plants and animals have SOD present as one of the essential body constituents. However, as discussed previously, orally administered SOD does not work in the body. Therefore, this reference adds nothing to the previously discussed Ginoux et al reference.

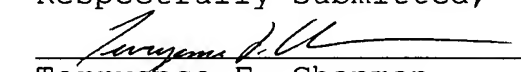
The Postaire et al reference discloses pharmaceutical compositions containing a superoxide dismutase and at least one lipid or protein. This reference discloses the use of SOD in the treatment of inflammatory processes, such as rheumatism and fibrosis, viral processes, such as HIV infection, and toxic conditions associated with the presence of substantial amounts of oxygen, such as central nervous system disorders,

ischemia, non-vascular gastrointestinal disorders, eye disorders or control of the undesirable effects of anti-cancer treatments. However, there is no disclosure in Postaire et al that the treated SOD disclosed there could be used in the treatment of cancerous tumors per se. Therefore, Applicants respectfully submit that Postaire et al in combination with the previously discussed references does not present a showing of prima facie obviousness under 35 USC 103 with respect to the presently claimed invention.

In order to further establish the patentability of the claimed invention over the prior art cited by the Examiner, Applicants are enclosing herewith a Declaration Under 37 CFR 1.132. As shown by the enclosed Declaration, the administration of a SOD/gliadin combination resulted in a more significant increase in SOD levels throughout the body as opposed to the administration of SOD melon extract alone. This is clearly unexpected in light of the prior art cited by the Examiner and establishes the patentability of the presently claimed invention thereover. The executed Declaration will be submitted to the Patent Office upon Applicants' receipt of same from Japan.

Reconsideration of the present application and the passing of it to issue is respectfully solicited.

Respectfully submitted,



TFC/smd

Terryence F. Chapman

FLYNN, THIEL, BOUTELL & TANIS, P.C.
2026 Rambling Road
Kalamazoo, MI 49008-1631
Phone: (269) 381-1156
Fax: (269) 381-5465

Dale H. Thiel	Reg. No. 24 323
David G. Boutell	Reg. No. 25 072
Ronald J. Tanis	Reg. No. 22 724
Terryence F. Chapman	Reg. No. 32 549
Mark L. Maki	Reg. No. 36 589
Liane L. Churney	Reg. No. 40 694
Brian R. Tumm	Reg. No. 36 328
Steven R. Thiel	Reg. No. 53 685
Donald J. Wallace	Reg. No. 43 977
Sidney B. Williams, Jr.	Reg. No. 24 949

Encl: References 1-5
Unexecuted Declaration Under 37 CFR 1.132
Replacement Abstract
Postal Card

References

Ref. 1. Huber W: Orgotein—(bovine Cu-Zn superoxide dismutase), an anti-inflamatory protein drug: discovery, toxicology and pharmacology. Eur J Rheumatol Inflamm. 1981;4(2):173-82.

Eur J Rheumatol Inflamm. 1981;4(2):173-82. Related Articles, Links

Orgotein--(bovine Cu-Zn superoxide dismutase), an anti-inflammatory protein drug: discovery, toxicology and pharmacology.

Huber W.

Orgotein is the generic name adopted by the USAN Council for drug versions of the Cu-Zn superoxide dismutases. It is obtained from bovine liver by a process sequentially involving heat treatment, enzymatic digestion of other proteins and purification to homogeneity by molecular sieve and ion-exchange chromatography. Orgotein occurs naturally in all mammalian cells, with liver, kidney, and erythrocytes being the richest sources. Prior to employing Orgotein in the clinic, a variety of toxicological and pharmacological investigations in animals have been conducted. The results of these studies are being presented. They indicate that Orgotein possesses a potent anti-inflammatory activity coupled with a pronounced lack of general pharmacological effects, and that its toxicity is of an extremely low order. Orgotein, a major topic of this workshop, is the generic name adopted in 1971 by the U.S. Adopted Names Council for drug versions of the Cu-Zn superoxide dismutases (SOD).

Ref. 2. . Zidenberg-Cherr S, Keen C, Lonnerdahl B et al: Dietary superoxide dismutase does not affect tissue levels Am J Clin Nutr 1983; 37(1):5-7.

Ref.3: Vouldoukis I. et al. Fc-receptor-mediated intracellular delivery of Cu/Zn-superoxide dismutase protects against redox-induced apoptosis through a nitric oxide dependent mechanism. Molecular Medicine 6:(12) 1042-1053, 2000.

Ref. 4

Med Hypotheses. 2000 Jul;55(1):29-35. Related Articles, Links

A threshold concept for cancer therapy.

Kong Q, Beel JA, Lillehei KO.

Department of Neurosurgery, University of Colorado Health Sciences Center, Denver 80262, USA. qing.kong@uchsc.edu

Oxygen free radicals have been implicated in many disease processes, including aging and carcinogenesis, and have been associated with a variety of complications resulting from the treatment of cancer. As a result, the treatment of free radical-induced disease with antioxidants or free radical scavengers has become an important therapeutic modality. Ironically, these same oxygen free radicals also play a critical role in anti-cancer therapies. The use of antioxidants such as superoxide dismutase (SOD), in this setting, has been found to decrease the efficacy of anti-tumor therapies, which depend on free radical generation for their action. In addition, increased antioxidant activity can often be utilized by the tumor cell to favor increased growth. Therefore, the appropriate application of oxygen free radicals and antioxidants seems to be critically important in designing proper strategies for both prevention and treatment of malignant disorders. This review will summarize free radical and antioxidant regimens that have been employed to date, examine some of the problems associated with these regimens, introduce the 'threshold concept' explaining the dual effects of oxygen free radicals and antioxidants, and discuss a novel hypothesis regarding therapy that could potentially improve outcome in cancer patients.

Ref. 5:

Br J Cancer. 2000 Oct;83(7):928-34. Related Articles, Links

Suppression of manganese superoxide dismutase augments sensitivity to radiation, hyperthermia and doxorubicin in colon cancer cell lines by inducing apoptosis.

Kuninaka S, Ichinose Y, Koja K, Toh Y.

Clinical Research Institute, Department of Chest Surgery, Gastroenterologic Surgery, National Kyushu Cancer Center, Notame 3-1-1, Minami-ku, Fukuoka, 811-1395, Japan.

Increased expression of manganese superoxide dismutase (Mn-SOD), one of the mitochondrial enzymes involved in the redox system, has been shown to diminish the cytotoxic effects of several anti-cancer modalities, including tumour necrosis factor-alpha, ionizing radiation, certain chemotherapeutic agents and hyperthermia. We asked if Mn-SOD is a potential target to augment the sensitivity of cancer cells to various anti-cancer treatments and for this we established stable Mn-SOD antisense RNA expressing cell clones from two human colon cancer cell lines, HCT116 (p53 wild-type) and DLD1 (p53 mutant-type). Suppression of Mn-SOD in HCT116 was accompanied by an increased sensitivity to radiation, hyperthermia and doxorubicin, as compared with findings in controls. The mitochondrial permeability transition, as measured by a decrease of the mitochondrial transmembrane potential was more intensely induced by radiation in HCT116 antisense clones than in the control, an event followed by a greater extent of DNA fragmentation. Apoptosis was also induced by hyperthermia more intensely in HCT116 antisense clones than in the control. On the other hand, DLD1 antisense clones did not exhibit any enhancement of sensitivity to any of these treatments. These data support the possibility that inhibition of Mn-SOD activity renders colon cancer cells with wild-type p53 susceptible to apoptosis induced by radiation, hyperthermia and selected anti-cancer drugs. Therefore, we suggest that Mn-SOD could be a target molecule to overcome the resistance to anti-cancer treatments in some colon cancer cells carrying wild-type p53.

PMID: 10970696 [PubMed - indexed for MEDLINE]